LANDMARK UNIVERSITY, OMU-ARAN

COURSE COMPACT

COLLEGE: SCIENCE AND ENGINEERING
DEPARTMENT: BIOLOGICAL SCIENCES
PROGRAMME: BIOCHEMISTRY
COURSE COMPACT for: ANALYTICAL BIOCHEMISTRY

Course

Course code:	BCH 314
Course title:	Analytical Biochemistry
Credit unit:	3
Course status:	Compulsory

Lecturers' Data

Name of the lecturers:	Dr. O.M. Oluba (Ph.D) Dr. (Mrs.) O.J. Awakan (Ph.D)
Department:	Biological Sciences
College:	Science and Engineering
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Consultation Hours: Wed – Fri, 1-2 pm

INTRODUCTION TO THE COURSE

Course Description:

In the era of multidisciplinary approach, the basic techniques in Biochemistry are much needed by the students of Biochemistry both at the undergraduate and postgraduate levels. This course covers various analytical and experimental protocols applicable in diverse areas of biochemical research. It discusses all the relevant biochemical techniques like spectroscopy, chromatography, electrophoresis, immunochemistry, radiolabelling, manometry, ultrafiltration, X-ray diffraction, centrifugation etc.

Course Justification:

The focus of the course lies in introducing analytical techniques applicable in the context of biomedical/clinical projects. It also teaches undergraduate biochemistry students instrumental bio-analytic chemistry required for biochemical investigations.

The course uniquely integrates the theories and practices that drive the various fields of biochemistry and comprehensively covers key analytical techniques that underpin recent biochemical advances and discoveries.

Course Objectives:

The course is aimed at exposing students in the biochemistry programme to the basics of instrumentation and bio-analytical techniques. At the end of the course, students would be able to;

i. Describe the working principles of the various instrumentation and analytical techniques
ii. Identify common applications, strengths and limitations of the various biochemical techniques.
iii. Describe the analytical techniques applicable in the analysis of biologic materials
iv. Demonstrate relevant skills in the application of the various analytical techniques to resolve or analyse biochemical problems.

Course Content:

Principles of instrumentation and analytical techniques; Manometry; Spectroscopy; Ultrafiltration; Chromatography; Electrophoresis; Centrifugation; X-ray diffraction; Immunochemical techniques; Viscosity measurements; Autoradiology and techniques of radiolabelling; (Laboratory practical should include areas of interest to academic staff to cut across a wide spectrum of general Biochemistry).

Course Expectations:

Attendance and full participation in Class is expected from all students.

S/N	GRADING	SCORE (%)
1.	Continuous Assessments • C.AI • C.AII (Mid-Semester Test) • C.AIII	 7% 15% 8%
2.	Assignment	
3.	Practical (Laboratory work)/ Case Studies	10%
4.	Final Examination	60%
5.	Total	100

Course Delivery Strategies:

Course delivery will be by electronic method, giving explanation and establishing scientific facts, while creating inquisitiveness in the minds of the students. Assignments will be given out to students periodically either individually or in groups.

Course Duration:

Three hours per week for 15 weeks (45hours)

LECTURE CONTENT

Module 1

Week 1: Introduction to the principles of instrumentation and analytical techniques

Objective: The students at the end of the lectures for the week should be able to:

(i) Describe how to select suitable analytical technique for use
(ii) Explain "qualitative analysis" and "quantitative analysis"
(iii) Explain how to ensure data reliability

Description: First hour: Explain what influences the choice of analytical techniques for a particular sample
Second hour: Explain "qualitative analysis" and "quantitative analysis"
Third hour: Ensuring data reliability

Study Questions:

1. What factor(s) do you consider in the selection of a suitable analytical technique in the separation of a sample?
2. How do you ensure data reliability?

Recommended reading

1. Modern Experimental Biochemistry. Third edition, Rodney Boyer
2. Modern Analytical Chemistry. (2000).David Harvey, McGraw Hill Higher Education
3. Analytical Biochemistry. Third Edition (1998), David Holme and Hazel Peck.
4. Biochemistry, Third edition (2005) by Voet and Voet, Wiley, ISBN: 978-0-471-19350-0
5. Harper's Illustrated Biochemistry, (2003). Twenty-sixth edition. McGraw-Hill companies limited. ISBN-0-07-121766-5
6. Nelson, D. L. and Cox, M. M. (2004) Integration and Hormonal Regulation of Mammalian Metabolism. Lehninger Principles of Biochemistry. 4th edition. Worth Publishers, New York

Module 2

Weeks 2&3: Spectroscopy

Objective: The students at the end of the lectures for the week should be able to:

i. Explain the electromagnetic emission spectrum.
ii. Explain the theory of light absorption and transmission (Beer-Lamber's law).
iii. Draw a schematic diagram of a UV/visible spectrophotometer (power supply, sight sources, monochromators, detector and measuring device).
iv. Identify the five main sections of spectrophotometer (radiation source, monochromator, photometer, sample area and detector area).
v. Describe the types of light sources, cells regions of the spectrum covered by visible and UV spectrophotometer.
vi. Describe the operation of the spectrophotometer.
vii. Describe the transmissive power of solvents (against distilled water).
viii. Describe the absorbance characteristics of some compound, e.g potassium dichromate, Haemoglobin etc.

ix. Describe ultraviolet spectrum as plot of the wavelength or frequency of absorption versus the absorption intensity.

Description: First and second hour: Theory/principles of spectroscopy
Third and fourth hour: Spectrophotometry: Instrumentation
Fifth and sixth hour: Types and applications

Study Question:

1. In which of the following techniques is the Beer-Lambert's relationship of significance?
    a) Fluorescence
    b) Flame emission spectroscopy
    c) Atomic absorption spectroscopy
    d) Nuclear magnetic resonance spectroscopy

Recommended reading

1. Modern Experimental Biochemistry. Third edition, Rodney Boyer
2. Modern Analytical Chemistry. (2000).David Harvey, McGraw Hill Higher Education
3. Analytical Biochemistry. Third Edition (1998), David Holme and Hazel Peck.
4. Biochemistry, Third edition (2005) by Voet and Voet, Wiley, ISBN: 978-0-471-19350-0
5. Harper's Illustrated Biochemistry, (2003). Twenty-sixth edition. McGraw-Hill companies limited. ISBN-0-07-121766-5
6. Nelson, D. L. and Cox, M. M. (2004) Integration and Hormonal Regulation of Mammalian Metabolism. Lehninger Principles of Biochemistry. 4th edition. Worth Publishers, New York

Weeks 4 & 5: Centrifugation

Objective: The students at the end of the lectures for the week should be able to:

i. Define centrifugation
ii. Describe the various principles of centrifugation
iii. Explain the applications of centrifugation as a separation technique.
iv. List the parameters which determine the sedimentation rate
v. List the parameters which determine the successful separation of mixture of heterogeneous particles.
vi. Explain the importance of the density and viscosity of the medium in facilitating the separation.
vii. List the order of sedimentation of the major cell components.
viii. Define preparative centrifugation and analytical centrifugation and state the difference(s) between the two.
ix. List the applications of preparative and analytical centrifugation.
x. List the three major classes of preparative centrifugation (general purpose centrifuge, higher speed centrifugation and the preparative ultracentrifuge) their rotor designs and uses).

Description: First and second hour: Theory/principles of centrifugation
Third and fourth hour: Centrifugation: Instrumentation
Fifth and sixth hour: Centrifugation: Types and applications

Study Questions:

1. Describe how you would design a centrifuge experiment to isolate sediments containing cell nuclei.

2. Cytochrome C has an s value of 1 x 10^{-13} second and haemoglobin an s value of about 4.5 x 10^{-13} second. Which protein has the larger molecular weight?

Recommended reading

1. Modern Experimental Biochemistry. Third edition, Rodney Boyer
2. Nelson, D.L. and Cox, M.M. (2004). Lehninger Principles of Biochemistry. 4th edition. Worth Publishers, New York

Weeks 6 & 7: Electrophoresis

Objectives: The students at the end of the lectures for the week should be able to:

i. Define electrophoresis as a method of separation and identification based on movement of charged molecules in an electric field.
ii. Explain the principles/theory of electrophoresis.
iii. Explain the factors that govern the behaviour of charged particles in an electric field.
iv. Describe the effect of the mobility particles (e.g. pH, osmotic flow, diffusion, etc.) during electrophoresis.
v. Describe the effect of the intensity of an electric field, power supply, current type of buffer used etc. on electrophoretic run.
vi. Demonstrate electrophoretic run in gel.
vii. Draw the circuit diagram of an electrophoresis power supply.
viii. Describe the precautionary measures adopted in high voltage electrophoresis.
ix. Describe and apply methods of sample application and markers/standards in electrophoresis.
x. Detect and estimate sample components.
xi. Identify in electrophoresis the different types of support media.
xii. Prepare different support media.
xiii. Describe the different types of electrophoresis e.g. high voltage, moving boundary, iso-electro focusing etc.
xiv. List the application of electrophoresis.

Description: First and second hour: Theory/principles of electrophoresis
Third and fourth hour: Electrophoresis: Instrumentation
Fifth and sixth hour: Electrophoresis: Types and applications

Study Questions:

1. What physical characteristics of a biomolecule influence its rate of movement in an electrophoresis matrix?
2. Explain the purpose of each of the chemical reagents that are used for PAGE
    a. Acrylamide
    b. N-N' methylene bis-acrylamide
    c. TEMED
    d. Sodium dodecyl sulphate
    e. Coomasie blue dye
    f. Bromophemol blue

Recommended reading

1. Modern Analytical Chemistry. (2000).David Harvey, McGraw Hill Higher Education
2. Modern Experimental Biochemistry. Third edition, Rodney Boyer
3. Biochemistry, Third edition (2005) by Voet and Voet, An introduction to

electrophoresis. Wiley, ISBN: 978-0-471-19350-0

Week 8: Mid-semester test

Weeks 9 & 10: Chromatography

Objective: The students at the end of the lectures for the week should be able to:

i. Describe the basic principles of chromatography
ii. Mention the various types of chromatography techniques and their applications
iii. Explain the principles for each of the types of chromatography techniques

Description: First hour: An introduction to chromatography, partition vs adsorption chromatography, planar chromatography (paper and thin layer chromatography)

Second hour: General theory of column chromatography, Gas Chromatography, High performance liquid chromatography (qualitative and quantitative analyses)

Third hour: Liquid-solid adsorption chromatography, ion exchange chromatography, size exclusion chromatography, supercritical fluid chromatography

Study Questions:

1. The analysis of trihalomethanes in drinking water was carried out. A single standard gives the following results.

Trihalomethane	Concentration (ppb)	Peak Area
$CHCl_3$	1.30	1.35×10^4
$CHCl_2Br$	0.90	6.12×10^4
$CHClBr_2$	4.00	1.71×10^4
$CHBr_3$	1.20	1.52×10^4

Analysis of water from a drinking fountain gives areas of 1.56×10^4, 5.13×10^4, 1.49×10^4, and 1.76×10^4 for $CHCl_3$, $CHCl_2Br$, $CHClBr_2$, and $CHBr_3$, respectively. Determine the concentration of each of the trihalomethanes in the sample of water.

Recommended reading

1. Modern Analytical Chemistry. (2000).David Harvey, McGraw Hill Higher Education
2. Modern Experimental Biochemistry. Third edition, Rodney Boyer

Week 11: Immunochemical techniques

Objective: The students at the end of the lectures for the week should be able to:

i. Describe immunochemical techniques and principles
ii. Mention the various types of immunochemical techniques and applications

iii. Give an highlight of the pituitary hormones and their physiologic roles

Description: First hour: General processes of the immune response
Second hour: Antigen-antibody reactions
Third hour: Analytical techniques-precipitation reactions, analytical techniques-immunoassay

Study Questions:

1. Which of the following describes an IgG immunoglobulin
   a. It is a pentamer
   b. It has two antigen binding sites
   c. It has an RMM of 1.0×10^{-6} Dalton
   d. It is the major immunoglobulin of the blood

Recommended reading

1. Analytical Biochemistry. Third Edition (1998), David Holme and Hazel Peck.
2. Harper's Illustrated Biochemistry, (2003) Twenty-sixth edition. McGraw-Hill companies limited. ISBN-0-07-121766-5

Week 12: Autoradiology and techniques of radiolabelling

Objective: The students at the end of the lectures for the week should be able to:

i. Describe autoradiology and principles
ii. Explain the radiolabelling technique
iii. Highlight the relevance of techniques and applications

Description: First hour: Origin and properties of radioactivity, Biochemical uses of isotopes, detection and measurement of radioactivity
Second hour: application of radioisotopes, the technique of autoradiography
Third hour: Radioisotopes and safety

Study Questions:

Recommended reading

1. Analytical Biochemistry. Third Edition (1998), David Holme and Hazel Peck.
2. Modern Experimental Biochemistry. Third edition, Rodney Boyer

Week 13: Manometry

Objective: The students at the end of the lectures for the week should be able to:

i. Explain the principle and application of manometric Technique.
ii. Explain the Similarities and differences between manometric and the oxygen electrode.
iii. Define the terms Respiratory Quotent and Metabolic Quotent
iv. Explain the three types of manometry viz: Constant volume manometry; Constant Pressure manometry; Differential manometry
v. Describe the operation, and principle and calibration of the Warburg constant volume manometre.
vi. Describe the Gilson Differential Respirometre
vii. List the general procedure for the operation of a manometre

viii. List the application of manometry.

Description: First hour: Principles and applications of manometry
Second hour: Manometric techniques
Third hour: Instrumentation

Study Questions:

Recommended reading

1. Analytical Biochemistry. Third Edition (1998), David Holme and Hazel Peck.
2. Modern Experimental Biochemistry. Third edition, Rodney Boyer

Week 14: Viscosity measurement

Objective: The students at the end of the lectures for the week should be able to:

i. Describe the term viscosity
ii. Explain how to measure viscosity in the laboratory or the field
iii. Highlight the relevance of viscosity measurement
iv. Explain the difference between "dynamic" and "kinematic" viscosity measurement
v. Mention and explain the types of viscometers or rheometers

Description: First hour:
Second hour:
Third hour:

Study Questions:

Recommended reading

1. Analytical Biochemistry. Third Edition (1998), David Holme and Hazel Peck.
2. Modern Experimental Biochemistry. Third edition, Rodney Boyer

Week 14: X-ray diffraction

Objective: The students at the end of the lectures for the week should be able to:

1. Describe x-ray diffraction as a biochemical technique
2. Explain the principles and application areas
3. Mention merits and demerits of the technique

Description: First hour:
Second hour:
Third hour:

Study Questions:

Recommended reading

1. Analytical Biochemistry. Third Edition (1998), David Holme and Hazel Peck.
2. Modern Experimental Biochemistry. Third edition, Rodney Boyer

Week 15: Revision

Objective: The students at the end of the lectures for the week should be able To express all that they have learnt

HOD's COMMENTS: _____

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Name: _____ Signature _____ Date: _____